



Cocaine and heroin in waste water plants: A 1-year study in the city of Florence, Italy

Francesco Mari^{a,*}, Lucia Politi^a, Annibale Biggeri^{b,c}, Gabriele Accetta^c, Claudia Trignano^d, Marianna Di Padua^a, Elisabetta Bertol^a

^a Division of Forensic Toxicology, Department of Anatomy, Histology, and Legal Medicine, University of Florence, Viale Morgagni, 85, 50134 Florence, Italy

^b Department of Statistics "G. Parenti", University of Florence, Italy

^c Biostatistics Unit, ISPO Institute for Cancer Prevention, Florence, Italy

^d Institute of Legal Medicine, University of Sassari, Italy

ARTICLE INFO

Article history:

Received 17 December 2008

Received in revised form 9 April 2009

Accepted 16 April 2009

Available online 20 May 2009

Keywords:

Waste water

Illicit drugs

1-year study

ABSTRACT

The diffusion and trends in use of each substance is a basic information in policy planning of strategies aiming at deterrence of drug abuse or in the organization of the fight against drug trafficking. The actual diffusion of illicit drugs in a population is hardly measurable, but, among the various measures available, the analysis of waste water plants represents one of the most reliable source of data.

We analyzed waste water in order to monitor illicit drug use by local population. We investigated the use of cocaine and heroin in the city of Florence, Italy, over a 1-year (July 2006–June 2007) period using state-of-the-art measuring techniques from waste water samples.

Cocaine, benzoylecgonine, and morphine were determined in water samples by gas chromatography–mass spectrometer, and the amount of illicit substance was estimated.

Data indicate for cocaine a bimodal distribution (December and March), while heroin showed a main peak in April. The heroin-to-cocaine use ratio in terms of estimated doses per month ranged from 0.11 to 0.76, representing new evidence of wider distribution of cocaine than heroin in Florence.

Waste water analysis can become a valuable tool in monitoring use of illicit drugs over time. In particular, it can highlight changes in the magnitude and relative use of illicit drug at a population level thereby becoming useful to develop strategies against drug trafficking and abuse. If routinely performed, it can be part of Epidemiologic Surveillance Programmes on drug abuse.

© 2009 Elsevier Ireland Ltd. All rights reserved.

1. Introduction

The use of illicit drugs is an international problem and leads to increased morbidity, mortality, and healthcare costs. In Italy, for example, 5.3% of the population have used cocaine at least once in their life while 2.5% have used heroin [1]; these percentages are rising [2]. Italy spends more than a hundred million euro per year to combat illicit drug use, money that cannot be spent on education, infrastructure, or improving other aspects of the country.

To understand the actual diffusion of illicit drugs in a population is a difficult task. The population of drug users is elusive and any single source of ascertainment could underestimate the population size (e.g. emergency room visits, death by drug overdose, policy records, out-patient clinical record or health service data). Population-based surveys are flawed by under-

reporting. Capture-mark-recapture methods [3] have been proposed to this purpose but there is debate about limitations, e.g. Biggeri [4]. The main problems are the varying propensity of the individuals to be listed in a given source and modeling multiple sources dependence [5,6].

In any case, such methods mostly apply to problematic drug consumption or abuse, since the available data list derive from hospital, health agencies or policy records. The diffusion of non-problematic consumption is therefore not addressed here (see Ref. [7] and for an attempt to use community survey and administrative data see Ref. [8]).

We have approached the task of quantifying the diffusion of illicit drug use in the community differently. Among the various measures available to track drug abuse the analysis of waste water plants (WWPs) can represent one of the most reliable source of data. Indeed, WWPs analysis has been increasingly used in recent years to better understand the diffusion of illicit drug use [9–15]. Furthermore, the so-called sewage epidemiology has recently gained the interest of the European Monitoring Centre for Drugs and Drug Addiction [16]. While several studies have developed and

* Corresponding author. Tel.: +39 055 7947208; fax: +39 055 7947208.

E-mail address: francesco.mari@unifi.it (F. Mari).

validated methods for drug detection in waste water, none of these evaluations – to the best of our knowledge – included a longitudinal evaluation of an entire city. In this perspective this approach to directly estimates the burden of illicit drug use could be complementary or even alternative to capture–recapture designs.

Currently many analytical methods are present in the literature and data published from a number of Countries or cities are potentially of great interest. However, factors influencing drug concentration in the water (e.g. flow and design of the water system, water treatments underwent, waste water temperature and pH) make comparisons among studies extremely complex. On the contrary, monitoring drug abuse patterns with periodic determination of a compound in the WWP within a single city can be of important meaning. Actually, the diffusion and trend use of each substance and the relationship among drugs in terms of the diffusion and trend, is a basic information in policy planning of strategies aiming at drug abuse deterrence or in the organization of the fight against drug trafficking. Such information can be helpful in focusing resources and expenditures toward a specific drug dealing or, if on routine, would be of undeniable value as an immediate indicator of the increment of use/abuse of a compound or class.

We investigated the use of cocaine and heroin in the city of Florence, Italy, over a 1-year period (July 2006–June 2007) using state-of-the-art measuring techniques from samples collected in waste water.

2. Materials and methods

WWPs samples were periodically collected, then specimens were submitted to a clean-up procedure in order to isolate the analytes of interest from interfering compounds and finally analyzed by gas chromatography–mass spectrometry.

2.1. Water samples

Two WWPs samples (1.5 L each) were collected on every first Monday of the month from July 2006 to June 2007 at each of two collection points of the water system serving the whole city of Florence and located at the banks (left and right) of the Arno river. Collection was performed before any physical or chemical treatment with the exception of the sedimentation and due to the sedimentation procedure lasting for 24 h samples are representative of a whole day. All sewage waters of the city flow at these collection points. The water flow was recorded by the water system flowmeter at each collection. Samples were stored at 4 °C and submitted to analysis within 3 days.

2.2. Standards and reagents

All pure standards (cocaine (COC), benzoylecgonine (BE), morphine (MO), and nalorphine), and N,O-bis-(trimethylsilyl)trifluoroacetamide (BSTFA) with 1% trimethylchlorosilane were purchased from Sigma (St. Louis, MO). All other reagents were purchased from J.T. Baker (Deventer, Holland).

2.3. Sample treatment and analysis

Samples were filtered on paper filters (Whatman Grade 1, 11 µm, Whatman, UK) and 0.5 µg (50 µl of the 10 ng/µl solution) of nalorphine (internal standard) was added. Each aliquot was solid phase extracted using Bond Elut Certify LRC cartridges (Varian, Harbor City, CA) and the procedure for basic drugs proposed by the manufacturer. The alkaline eluate was evaporated to dryness under nitrogen stream at room temperature and derivatized with 50 µl of BSTFA with 1% TMCS.

One microliter of the solution was injected in the gas chromatography–mass spectrometer (GC–MS, Thermoquest Trace GC/Finnigan Polaris Q MS) equipped with a 12 m × 0.2 mm I.D. phenylmethylsilicone 5% (5 MS) capillary column with a 0.33 µm film thickness. Carrier gas (helium) was kept constant at 1 mL/min. The injector was set at 300 °C and splitless injection was employed with split valve off time of 3 min. The column oven temperature was programmed to rise from an initial temperature of 60 °C hold for 0.5 min, increase to 130 °C at 10 °C/min, then to 300 °C at 40 °C/min, and hold at 300 °C for 3 min. For identification purposes, four ions were monitored for each analyte: *m/z* 82, 182, 272, 303 for COC (*m/z* 182 used for quantification); 82, 240, 346, 361 for BE (quantifier: 82); 324, 401, 414, 429 for MO (quantifier: 429); and 324, 414, 440, 455 for nalorphine (quantifier: 455).

A 4-point calibration curve was built at 25, 50, 100, and 200 ng/L for COC and BE and at 4.4, 8.8, 17.7, 35.3 ng/L for MO; quality control points were injected daily

with the calibration curve at 25, 50, 150 ng/L for COC and BE and at 3.7, 6.2, 25.5 ng/L for MO. Linearity, precision (as relative standard deviation, RSD), and accuracy (as bias) were evaluated by injecting calibration curve and quality control points during the same analysis sessions of the samples. Analyte recoveries were measured at 50 and 150 ng/L for COC and BE and for 6.2 and 25.5 ng/L for MO. HPLC-grade water (Fluka, St. Louis, MO) was used for the preparation of calibration and control points.

2.4. Validation

Commercially acquired HPLC-grade water did not produce any peaks at the retention times of the analytes or of the internal standard. The method was found to be linear over the whole range for the three analytes with correlation coefficients better than 0.9957. Precision and accuracy were always acceptable (better than 16% in RSD and bias percent at the lowest quality control and better than 15% at the other quality controls). Hence, the lowest quality control point (25 ng/L for COC and BE, 3.7 ng/L for MO) was accepted as lower limit of quantification for the three substances. Recoveries were in the following ranges: 87–103% (COC), 91–103% (BE), and 96–109% (MO).

2.5. Statistical analysis

Using data from the concentrations (ng/L) in water and from the water flow (L/day) COC, BE and MO loads (grams per day, g/day) were computed on each sampling site and each month. We also assessed the loads of total COC in water per day, adding the loads of COC and the loads of BE taking into account that the molar mass ratio between COC and BE is 1.048 (303.35/289.33). Similarly from MO to heroin we used the molar ratio (369.41/285.34).

Reliability of water concentration measurements was assessed by intraclass correlation coefficient from mixed effect ANOVA model [17].

The right bank of the river had a mean flow rate of 1657 liters per second (L/s). For the left bank flow rate was estimated as 90% of the right bank flow rate, based on expert opinion.

Water flows varied by season during the year. In Mediterranean climate precipitations are restricted to the spring period between February and June. We estimated the water flow by month using a James-Stein estimator which takes into account within and between season variability of water flow measurements [18]. This estimator smoothes the estimated pattern by months producing a more stable estimate of monthly water flow. For example the measured water flow for August was an outlier. To control for outlying observations we used a shrinkage estimator which takes into account of the overall seasonal pattern of water flows. Algebraically the James-Stein estimator is a weighted average between the monthly measurement and the global average. The amount of shrinkage is controlled by the weight, which is proportional to the variance ratio, between season over the total variance (the sum of between and within season variance). The larger the within season variance component the greater the shrinkage because this implies high sampling variability within season. The variance of the James-Stein estimator is in Ref. [19].

There was uncertainty in the left bank flow rate estimate, since this was based on expert opinion. We performed a sensitivity analysis using two different prior distributions. First, we carried out a numerical integration of the flow rate estimates assuming a Gaussian prior with maximum probability density at 90% and minimum at 40% (scheme A). Scheme A is based on a conservative scenario: conservative in the sense that values near our initial estimate of 90% are highly plausible. Second, the robustness of our loads estimate was further verified through an alternative prior (scheme B) for the left bank flow rate with maximum probability density at 40% and minimum at 90%. Scheme B describes a situation where our initial estimate of 90% is little plausible [20].

Monthly loads (g/day) for each substance considered were calculated multiplying concentrations by water flows. We specified a Generalized Linear Model for a Gamma distributed response variable (which is standard for modeling concentrations) with identity link (to preserve additivity in load calculations). Water flows estimates were introduced as an offset term. Since water flows are subjected to random variability we specified analytic weights equal to the inverse of the variance of the James-Stein estimates of water flow. Indicator variables for month, site and interaction terms were specified in the linear predictor. The model was fitted by maximum likelihood using STATA 10 [21,22].

All monthly load forecast and confidence intervals were based on model output.

3. Results

3.1. Estimation of drug concentration in WWP

Table 1 reports the concentrations (ng/L) of COC, BE, and MO in water from the right and left banks of Arno river, sampled during the period July 2006–June 2007 in Florence, Italy. COC, BE, and MO replicates at each sampling occasion are reported. The coefficient of reliability was 0.99 for COC, 0.98 for BE and 0.97 for MO.

Table 1

Concentration (ng/L) of cocaine, benzoylecgonine and morphine in water from the two collection points, July 2006–June 2007, Florence, Italy.

Period	Cocaine (ng/L)				Benzoylecgonine (ng/L)				Morphine (ng/L)			
	Right bank		Left bank		Right bank		Left bank		Right bank		Left bank	
	1st sample	2nd sample	1st sample	2nd sample	1st sample	2nd sample	1st sample	2nd sample	1st sample	2nd sample	1st sample	2nd sample
2006												
July	57.1	56.5	83.5	79.8	135.8	132.3	171.3	173.8	5.4	4.5	4.5	4.3
August	33.2	33.1	56.5	50.6	122.3	127.5	106.4	103.2	5.8	5.3	6.4	7.5
September	31.6	28.1	76.4	74.9	89.5	86.0	157.2	154.0	4.5	4.1	9.7	9.8
October	57.2	56.2	89.5	87.0	112.4	114.9	109.2	106.5	4.6	5.1	7.7	7.2
November	41.0	38.8	55.0	50.7	169.3	151.4	204.2	180.9	10.6	9.1	10.9	9.9
December	59.0	57.3	78.9	77.9	183.4	189.4	206.4	214.0	11.4	12.9	23.7	22.7
2007												
January	33.8	29.2	54.6	50.5	97.9	97.7	173.7	158.5	14.4	16.1	19.7	15.7
February	26.2	29.7	32.6	29.6	80.1	81.1	89.6	93.7	13.3	13.4	16.5	16.8
March	55.1	58.8	62.0	61.6	137.6	133.6	140.0	143.4	16.5	15.6	18.7	18.3
April	36.2	35.0	38.1	35.5	88.3	82.1	94.4	90.3	18.3	18.7	19.2	19.9
May	37.9	37.0	36.1	35.0	96.8	99.7	99.6	98.3	15.2	14.7	18.3	15.8
June	38.0	34.7	47.0	46.0	99.0	98.4	118.8	121.3	12.1	12.0	12.6	12.9

Table 2

Estimated loads (g/day) and 95% confidence interval of morphine and total cocaine in the WWP, Florence, Italy, July 2006–June 2007.

Month	Heroin (g/day)			Total cocaine (g/day)		
	Loads	95% IC		Loads	95% IC	
2006						
July	1.60	1.25	1.95	60.29	47.37	73.22
August	2.72	2.14	3.30	55.17	43.20	67.15
September	2.27	1.77	2.77	45.14	35.32	54.96
October	2.07	1.63	2.52	49.65	38.96	60.34
November	3.46	2.71	4.21	60.80	47.74	73.85
December	5.95	4.66	7.24	72.64	57.02	88.26
2007						
January	5.61	4.41	6.82	47.01	36.88	57.14
February	6.14	5.25	7.03	38.02	32.48	43.57
March	7.08	6.05	8.11	65.06	55.53	74.59
April	7.83	6.69	8.97	41.08	35.06	47.09
May	6.58	5.62	7.54	44.52	37.97	51.07
June	5.10	4.36	5.85	49.40	42.22	56.59

COC, BE, and MO loads (g/day) were computed on each sampling site and each month and reported in Table 2. Total COC peaked on November/December and March showing a bimodal distribution. Heroin showed a main peak in April (Fig. 1).

However there is uncertainty in expert opinion on the left bank flow rate. We reported in Table 3 the sensitivity analysis. Recall that left bank flow rate best expert estimate is 90% of right bank flow rate. Reductions under 40% were considered implausible. Therefore, in the first scenario, we assume a Gaussian weighting scheme with maximum weight for a coefficient of reduction of 90% and minimum for a reduction of 40% (scheme A). This is a conservative scenario because values near our best experts' estimate of 90% are highly probable. In the second scenario, we use an alternative weighting scheme (B) for the left bank flow rate with maximum weight at 40% and minimum at 90%. Scheme B describes a reverse situation where left bank flow rate is expected to be much less than right bank. The overall load is reduced by 9% under scheme A and by 19% by scheme B.

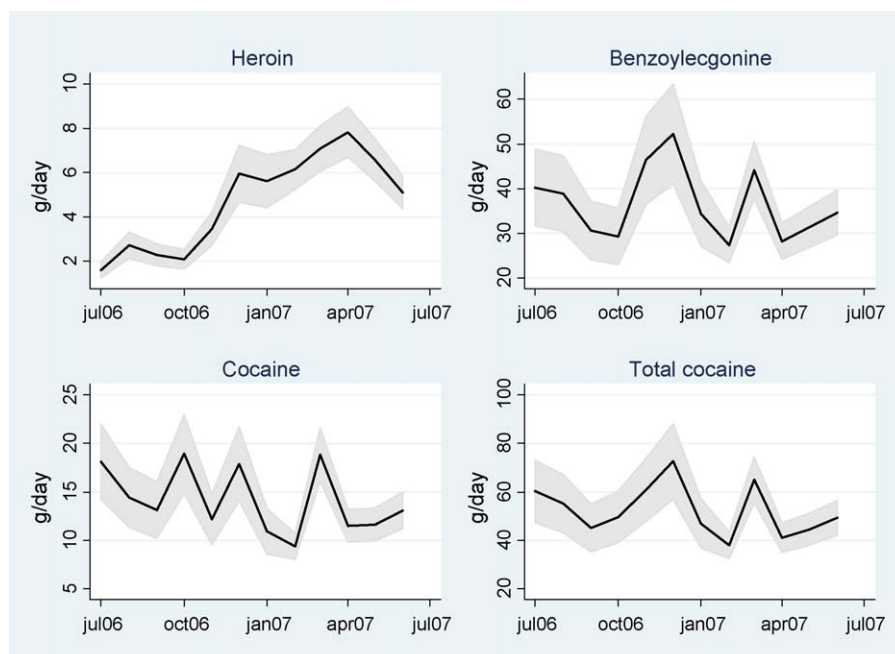
**Fig. 1.** Loads (g/day) and 95% confidence intervals of morphine, benzoylecgonine, cocaine and total cocaine in the WWP, Florence, Italy, July 2006–June 2007.

Table 3

Sensitivity analysis with respect to the left bank flow rate. Loads estimate (g/day), under two different weighting schemes. Schemes A and B are based on a Gaussian distribution with mean at 90% and 40%, respectively. Variance is the same in both schemes, with probability values less than 40% under scheme A equal to 0.01. Both weight schemes are truncated below 40% and beyond 90%.

Month	Loads estimate (g/day)			
	Scheme A		Scheme B	
	Heroin	Cocaine	Heroin	Cocaine
2006				
July	1.48	54.80	1.32	47.53
August	2.48	50.82	2.16	45.05
September	2.02	40.33	1.68	33.95
October	1.87	45.44	1.61	39.86
November	3.18	55.47	2.80	48.42
December	5.32	66.39	4.49	58.12
2007				
January	5.14	42.27	4.50	35.99
February	5.60	34.82	4.88	30.57
March	6.48	59.76	5.68	52.74
April	7.19	37.71	6.35	33.25
May	6.02	41.01	5.29	36.37
June	4.69	45.06	4.14	39.31

4. Discussion

COC, BE, and MO were detected in all specimens collected every month from both banks serving all the city of Florence, throughout one entire year. Hence, differently from all previous publications, which contemplated a single sampling at a single collection site, for the first time the concentration of the three compounds was estimated for one entire city over a 12-month period.

The MO loads permitted to extrapolate the amount of heroin used by calculating heroin equivalents to MO concentration. Heroin extrapolation might be affected by therapeutic MO use in cancer pain management. However correlation between MO loads and monthly number of deaths due to cancer diseases showed a negligible association (Spearman correlation coefficient = 0.007; details not shown).

Zuccato et al. [23] suggested to calculate COC by multiplying BE concentration by 2.33. This factor takes into account cocaine-to-benzoylgonine molar mass ratio (1.05) and the average molar fraction (45%) of a COC dose that is excreted as BE [24]. Here, COC was estimated by adding BE transformed into COC equivalents to COC. This calculation has the drawback that potentially could consider also COC thrown into waste water system, for example during a police pursuit. However, for the whole period considered, our calculation resulted in lower results than that suggested by Zuccato, hence, it is unlikely that it overestimates COC use by including wasted COC. The ratio between BE and COC concentration was consistent with the literature [11] and relatively constant. The ratio between the COC consumption and the total COC load in water ranges from 1.25 to 1.80 with a mean of 1.62. In consideration of the fact that many are the factors impacting the concentration of drugs in waste water and many of these do not undergo our control, it is not impossible that the correction factor of 2.33 applied to benzoylgonine is overestimating COC use. In fact, the factor is based on the average of 45% of COC transformed into BE. These data are of course sound and reliable, but referred to the first 24 h after intake [24]. Since in waste water we are virtually detecting BE from COC intake “to infinity”, it is rational to hypothesize that BE in water represents a higher percentage than 45%. On the basis of all these considerations the sum of COC and BE equivalents was used for further calculations. However, it is likely that we are still underestimating the amount of all substances in WWP, for a number of reasons. For example, it has been recently demonstrated that COC itself is unstable at 20 °C and pH 6 [11]. Furthermore, we are aware of possible weekly variation in the

consumption. Our estimates are therefore more imprecise being based only on 1 day per month. Moreover our data suggest monthly variation being greater than weekly variation [25].

As shown in Fig. 1, heroin use seems to be at its maximum during the spring and, even though this is not of great reliability, an increment in police seizures of heroin was observed in the same period. Moreover, both heroin and COC seem to show a peak in December and, considering that sampling was performed on the first Monday of each month, December sample will include New Years Eve.

The number of doses used in each month can be deducted considering the loads of heroin and COC and an average amount of 25 mg per dose for heroin and 100 mg for COC (mg/dose values were arbitrarily assessed on the basis of the laboratory experience in seizures analysis). The calculated number of doses per month ranged from 1984 (July) and 9396 (April) for heroin and from 10645.6 (February) and 26876.8 (December) for cocaine. The total number of doses estimated in the whole city during the considered period stands for a use of at least 68,322 doses of heroin and 192,884 doses of cocaine. The Annual report on drug addiction in Italy in 2005 [1] estimated that 1.3% of the whole Italian population used heroin at least once in their life and 6.7% used COC, and, in the last year, 0.3% used heroin and 2.2% used COC. In other words, 300,000 Italians use COC and 45,000 use heroin at least once a month and 32,000 use COC everyday and 20,000 use heroin everyday. According to these data the ratio between heroin and COC use is 0.15 for occasional use and 0.625 for everyday use. These ratios seem in good accordance with our results: in fact, the heroin to COC ratio in terms of estimated doses was from 0.11 to 0.76.

The results highlighted that, despite all the factors – either known or unknown – influencing the substance concentration in the water and the estimation of the magnitude of the use, WWP analysis over time can ascertain the trend of the use of illicit drug in a definite area. In particular, such evidence in changes of the magnitude of the diffusion of each substance and the ratio among different drugs will be valuable in planning the campaign against drug trafficking and abuse. Moreover, if continuously performed on routine, WWP analysis can be a prompt indicator of the rising/declining of a determined substance, thus helping focusing economic and human resources.

In conclusion, we have demonstrated that WWP analysis can be used to estimate the magnitude of drug use in an entire city. This study permitted to highlight that, in spite of the risk of taking into account “police pursuit” COC, the sum of measured COC and BE seems more reliable than BE transformed into taken COC. Moreover, the ratio between the heroin and COC use appears in accordance with the last Annual report on drug addiction in Italy, being a further confirmation of the decrease of the splitting of use between heroin and COC. While there are several factors that affect the precision of our estimates – such as randomness of sampling, uncertainty of flow rate, therapeutic use of morphine, and analyte stability – the continuous monitoring of waste water can be used to estimate illicit drug use in a population throughout the year. The analysis of waste water thus provides a valuable method for focusing economic and human resources in the fight against drug trafficking and abuse.

Conflict of interest

All authors disclose any financial or personal relationships with other people or organizations that could have inappropriately influenced this work.

Acknowledgment

This study was partially supported by a grant from the “Ente Cassa di Risparmio di Firenze”.

The authors wish to thank Maria Grazia Di Milia, M.Sc. for the precious technical support.

References

- [1] Annual Report on Drug Dependence in Italy in 2005. Ministero della Solidarietà Sociale. Relazione annuale al Parlamento sullo stato delle tossicodipendenze in Italia nel 2005.
- [2] European Monitoring Centre for Drugs and Drug Addiction. Annual Report on the State of The Drugs Problem in Europe 2007, European Union EMCDDA, Lisbon, 2008.
- [3] E.B. Hook, R.R. Regal, Capture–recapture methods in epidemiology: methods and limitations, *Epidemiol. Rev.* 17 (1995) 243–264.
- [4] A. Biggeri, Capture–recapture experiments in observational epidemiology. IBC Special Contributed Session “Capture–Recapture Procedures in Public Health”, in: Proceedings of the XXII International Biometrics Conference, Cairns, 2004.
- [5] E.B. Hook, R.R. Regal, Accuracy of alternative approaches to capture–recapture estimates of disease frequency: internal validity analysis of data from five sources, *Am. J. Epidemiol.* 152 (2000) 771–779.
- [6] R. King, S.H. Bird, S.P. Brooks, S.J. Hutchinson, G. Hay, Prior information in behavioral capture–recapture methods: demographic influences on drug injectors’ propensity to be listed in data sources and their drug-related mortality, *Am. J. Epidemiol.* 162 (2005) 694–703.
- [7] M.T. Brugal, A. Domingo-Salvany, E. Diaz de Quijano, L. Torralba, Prevalence of problematic cocaine consumption in a city of southern Europe using capture–recapture with a single list, *J. Urban Health.* 81 (2004) 416–427.
- [8] V.D. Hope, M. Hickman, K. Tilling, Capturing crack cocaine use: estimating the prevalence of crack cocaine use in London using capture–recapture with covariates, *Addiction* 100 (2005) 1701–1708.
- [9] S. Castiglioni, E. Zuccato, E. Crisci, C. Chiabrande, R. Fanelli, R. Bagnati, Identification and measurement of illicit drugs and their metabolites in urban wastewater by liquid chromatography–tandem mass spectrometry, *Anal. Chem.* 78 (2006) 8421–8429.
- [10] M. Huerta-Fontela, M.T. Galceran, F. Ventura, Ultraperformance liquid chromatography–tandem mass spectrometry analysis of stimulatory drugs of abuse in wastewater and surface waters, *Anal. Chem.* 79 (2007) 3821–3829.
- [11] A. Gheorghe, A. van Nuijs, B. Pecceu, L. Bervoets, P.G. Jorens, R. Blust, H. Neels, A. Covaci, Analysis of cocaine and its principal metabolites in waste and surface water using solid-phase extraction and liquid chromatography–ion trap tandem mass spectrometry, *Anal. Bioanal. Chem.* 391 (2008) 1309–1319.
- [12] J. Bones, K.V. Thomas, B. Paull, Using environmental analytical data to estimate levels of community consumption of illicit drugs and abused pharmaceuticals, *J. Environ. Monit.* 9 (2007) 701–707.
- [13] M. Huerta-Fontela, M.T. Galceran, J. Martin-Alonso, F. Ventura, Occurrence of psychoactive stimulatory drugs in wastewaters in north-eastern Spain, *Sci. Total Environ.* 397 (2008) 31–40.
- [14] B. Kasprzyk-Hordern, R.M. Dinsdale, A.J. Guwy, Multiresidue methods for the analysis of pharmaceuticals, personal care products and illicit drugs in surface water and wastewater by solid-phase extraction and ultra performance liquid chromatography–electrospray tandem mass spectrometry, *Anal. Bioanal. Chem.* 39 (2008) 1293–1308.
- [15] E. Zuccato, S. Castiglioni, R. Bagnati, C. Chiabrande, P. Grassi, R. Fanelli, Illicit drugs, a novel group of environmental contaminants, *Water Res.* 42 (2008) 961–968.
- [16] European Monitoring Centre for Drugs, Drug Addiction, Assessing Illicit Drugs in Wastewater, Lisbon, European Union EMCDDA, 2008.
- [17] E. White, B.K. Armstrong, R. Saracci, Principles of exposure measurement in epidemiology: collecting, in: *Evaluating and Improving Measures of Disease Risk Factors*, 2nd ed., Oxford Press, 2008.
- [18] B. Efron, C. Morris, Data analysis using Stein’s estimator and its generalizations, *JASA* 70 (1975) 311–319.
- [19] A. Skrondal, S. Rabe-Hesketh, Generalized latent variable modeling: multilevel, in: *Longitudinal and Structural Equation Models*, Chapman & Hall/CRC Press, Boca Raton (FL), 2004.
- [20] D.J. Spiegelhalter, K.R. Abrams, J.P. Myles, Bayesian Approaches to Clinical Trials and Health-care Evaluation, John Wiley and Son, 2004.
- [21] StataCorp, Stata Statistical Software: Release 10, StataCorp LP, College Station, TX, 2007.
- [22] P. McCullagh, J.A. Nelder, Generalized Linear Models, 2nd ed., Chapman and Hall, New York, 1989.
- [23] E. Zuccato, C. Chiabrande, S. Castiglioni, D. Calamari, R. Bagnati, S. Schiarea, R. Fanelli, Cocaine in surface waters: a new evidence-based tool to monitor community drug abuse, *Environ. Health* 5 (2005) 4–14.
- [24] R.C. Baselt, Disposition of Toxic Drugs and Chemicals in Man, 7th ed., Biomedical Publications, Foster City (CA), 2004.
- [25] E. Zuccato, C. Chiabrande, S. Castiglioni, R. Bagnati, R. Fanelli, Estimating community drug abuse by wastewater analysis, *Environ. Health Perspect.* 116 (2008) 1027–1032.